PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 0077 WO 00 ORD	**FOR FURTHER ACTION**	See Notification of Transmittal of International Preliminary Examination Report (Form PCT/IPEA/416)
International application No. PCT/EP 03/06341	International filing date *(day/month/year)* 13.06.2003	Priority date *(day/month/year)* 13.06.2002
International Patent Classification (IPC) or both national classification and IPC C07K16/28		
Applicant CRUCELL HOLLAND B.V. et al		

1. This international preliminary examination report has been prepared by this International Preliminary Examining Authority and is transmitted to the applicant according to Article 36.

2. This REPORT consists of a total of 9 sheets, including this cover sheet.

   ☒ This report is also accompanied by ANNEXES, i.e. sheets of the description, claims and/or drawings which have been amended and are the basis for this report and/or sheets containing rectifications made before this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions under the PCT).

   These annexes consist of a total of 7 sheets.

3. This report contains indications relating to the following items:

   - I ☒ Basis of the opinion
   - II ☐ Priority
   - III ☒ Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
   - IV ☐ Lack of unity of invention
   - V ☒ Reasoned statement under Rule 66.2(a)(ii) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
   - VI ☐ Certain documents cited
   - VII ☐ Certain defects in the international application
   - VIII ☐ Certain observations on the international application

Date of submission of the demand	Date of completion of this report
06.01.2004	21.07.2004
Name and mailing address of the international preliminary examining authority: European Patent Office D-80298 Munich Tel. +49 89 2399 - 0 Tx: 523656 epmu d Fax: +49 89 2399 - 4465	Authorized Officer Lechner, O Telephone No. +49 89 2399-8687

INTERNATIONAL PRELIMINARY EXAMINATION REPORT

International application No. PCT/EP 03/06341

I. Basis of the report

1. With regard to the **elements** of the international application *(Replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report since they do not contain amendments (Rules 70.16 and 70.17))*:

Description, Pages

1-84 as originally filed

Claims, Numbers

1-28 received on 07.06.2004 with letter of 07.06.2004

Drawings, Sheets

1/21-21/21 as originally filed

Sequence listing part of the description, pages:

85-130, as originally filed

2. With regard to the **language**, all the elements marked above were available or furnished to this Authority in the language in which the international application was filed, unless otherwise indicated under this item.

   These elements were available or furnished to this Authority in the following language: , which is:

   ☐ the language of a translation furnished for the purposes of the international search (under Rule 23.1(b)).

   ☐ the language of publication of the international application (under Rule 48.3(b)).

   ☐ the language of a translation furnished for the purposes of international preliminary examination (under Rule 55.2 and/or 55.3).

3. With regard to any **nucleotide and/or amino acid sequence** disclosed in the international application, the international preliminary examination was carried out on the basis of the sequence listing:

   ☒ contained in the international application in written form.

   ☒ filed together with the international application in computer readable form.

   ☐ furnished subsequently to this Authority in written form.

   ☐ furnished subsequently to this Authority in computer readable form.

   ☐ The statement that the subsequently furnished written sequence listing does not go beyond the disclosure in the international application as filed has been furnished.

   ☐ The statement that the information recorded in computer readable form is identical to the written sequence listing has been furnished.

4. The amendments have resulted in the cancellation of:

   ☐ the description, pages:

   ☐ the claims, Nos.:

   ☐ the drawings, sheets:

5. ☐ This report has been established as if (some of) the amendments had not been made, since they have been considered to go beyond the disclosure as filed (Rule 70.2(c)).

   (Any replacement sheet containing such amendments must be referred to under item 1 and annexed to this report.)

6. Additional observations, if necessary:

III. Non-establishment of opinion with regard to novelty, inventive step and industrial applicability

1. The questions whether the claimed invention appears to be novel, to involve an inventive step (to be non-obvious), or to be industrially applicable have not been examined in respect of:

   ☐ the entire international application,

   ☒ claims Nos. 1, 2, 9, 10, 11, 15-28 (all in part)

   because:

   ☒ the said international application, or the said claims Nos. 9, 10, 27, 28 (concerning industrial applicability) relate to the following subject matter which does not require an international preliminary examination (specify):

   see separate sheet

   ☐ the description, claims or drawings *(indicate particular elements below)* or said claims Nos. are so unclear that no meaningful opinion could be formed *(specify)*:

   ☐ the claims, or said claims Nos. are so inadequately supported by the description that no meaningful opinion could be formed.

   ☒ no international search report has been established for the said claims Nos. 1, 2, 6, 9, 11, 12, 15-28 (all in part)

2. A meaningful international preliminary examination cannot be carried out due to the failure of the nucleotide and/or amino acid sequence listing to comply with the standard provided for in Annex C of the Administrative Instructions:

   ☐ the written form has not been furnished or does not comply with the Standard.

   ☐ the computer readable form has not been furnished or does not comply with the Standard.

V. Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Yes:	Claims	1-28
	No:	Claims	
Inventive step (IS)	Yes:	Claims	2
	No:	Claims	1, 3-28
Industrial applicability (IA)	Yes:	Claims	
	No:	Claims	see sep-sheet

2. Citations and explanations

see separate sheet

Item I

Amended **claims 1-28** filed with the letter dated 7.6.2004 do not introduce subject-matter which extends beyond the content of the application as filed and thus are considered to fulfil the requirements of **Article 34(2)(b) PCT**.

item III

1 **Non-establishment of opinion with regard to novelty, inventive step and industrial applicability (Rule 67.1, PCT)**

For the assessment of the present **claims 9** (as far as relating to human beings), **27, 28** on the question whether they are industrially applicable, no unified criteria exist in the PCT Contracting States. The patentability can also be dependent upon the formulation of the claims. The EPO, for example, does not recognize as industrially applicable the subject-matter of claims to the use of a compound in medical treatment, but may allow, however, claims to a known compound for first use in medical treatment and the use of such a compound for the manufacture of a medicament for a new medical treatment. **Claims 9, 27, 28** relate to subject matter considered by this Authority to be covered by the provisions of **Rule 67.1(iv), PCT**. Consequently, no opinion will be formulated with respect to the industrial applicability of the subject matter of these claims **(Article 34(4)(a)(i), PCT)**.

2 The examination of novelty and inventive step of **claims 1, 2, 6, 9, 11, 12, 15-28** was limited to those parts which have been searched, i.e. the use of anti-OX40 receptor antibodies and the molecules disclosed in **claims 3** and **4 (Rule 66.1(e), PCT)**.

3 **Major Clarity problems (Article 6, PCT)**

3.1 **Claim 10** does not meet the requirements of **Article 6, PCT** in that it is unclear as far as relating to a "cell derived from a human cell".

For the present examination of novelty and inventive step the claim was considered to relate to "host is a human cell".

3.2 **Claims 25, 26** are not acceptable under **Art. 6, PCT** (clarity). The therapeutic application is only functionally defined by a result to be achieved, i.e. enhancing the immune response in a human or animal and enhancing an immune response against a tumor, bacterial or viral antigen in a human or animal, respectively, which does not allow any practical application in the form of a defined, real treatment of a pathological condition (disease). The objection could be overcome by either introducing in the claims a list of pathological conditions (diseases) cited in the application, or by showing that means are available which would allow the skilled person to recognise

which additional condition(s) would fall within the functional definition. Consequently, the examination of novelty and inventive step for **claims 25** and **26** has been carried out interpreting the claims in view of the description (**Rule 66.1(e), PCT**), namely those parts relating to the specific diseases listed on page 57, §2 - 60, §1.

Item V

1 The following documents have been cited in the present written opinion; the numbering will be adhered to in the rest of the procedure:

D1 WO 99 42585 A (SISTERS OF PROVIDENCE IN OREGO) 26 August 1999 (1999-08-26)

D2 WEINBERG A D ET AL: 'Engagement of the OX-40 receptor in vivo enhances antitumor immunity' JOURNAL OF IMMUNOLOGY, THE WILLIAMS AND WILKINS CO. BALTIMORE, US, vol. 164, no. 4, 15 February 2000 (2000-02-15), pages 2160-2169, XP002224132 ISSN: 0022-1767

D3 US 2001/044522 A1 (GODFREY WAYNE ET AL) 22 November 2001 (2001-11-22)

D4 WEINBERG A D: 'OX40: targeted immunotherapy - implications for tempering autoimmunity and enhancing vaccines' TRENDS IN IMMUNOLOGY, ELSEVIER, CAMBRIDGE, GB, vol. 23, no. 2, 1 February 2002 (2002-02-01), pages 102-109, XP004347862 ISSN: 1471-4906

D5 VAUGHAN T J ET AL: 'Human antibodies by design' NATURE BIOTECHNOLOGY, NATURE PUBLISHING, US, vol. 16, June 1998 (1998-06), pages 535-539, XP002141139 ISSN: 1087-0156

2 Novelty (Article 33(2), PCT)

2.1 No documents are comprised in the known prior art explicitly disclosing human OX40 receptor agonistic binding antibodies, vectors in human host cells and the methods as disclosed in **claims 1-28.** Thus, the subject matter of **claims 1-28,** as far as examined (c.f. item III), appears to be novel in the sense of **Article 33(2), PCT.**

3 Inventive step (Article 33(3), PCT)

For the following reasons no inventive step in the sense of **Article 33(3), PCT** can be acknowledged for the subject matter of **claims 1, 3-28**:

D1 is considered to be the closest prior art and discloses the use of murine OX-40 agonistic Ab (Pharmingen Abs or Mab OX-86 obtainable from the ECACC) or of OX-40L for enhancing T-cell activation in vitro and the in vivo immune response to tumor cells

in humans. The Ab or ligands may be provided in nucleic acid form as well. Anti-OX40 binding agents are capable of stimulating T cells in vitro and to protect animals from tumors in vivo. Accordingly, the data suggest that OX-40R based therapy can generally enhance the immune system, not only for tumor immunity, but also as an immunologic adjuvant for all vaccine types e.g. also viral, bacterial, etc.. vaccines (c.f. p3, l 15 - p 4, l 16; p 5, l 35 - p 6, l 7, l 27-37; p 10, l 17 - p 13, l 11; claims 1-26).

3.1 claim 1, 3-28

The difference between **D1** and the subject matter of **claim 1**, as far as examined (c.f. item III), is the disclosure of human agonistic binding Ab capable of binding to and stimulating the human OX40 receptor.
The problem to be solved is to provide agonistic binding Ab capable of binding to and stimulating the human OX40 receptor with no intrinsic immunogenicity or toxicity.
The claimed solution is the use of human agonistic binding Ab.

The International Preliminary Examination Authority is still of the opinion, that in view of the teachings of **D1** and the fact that the human OX-40 amino acid sequence is known in the art (c.f **D3**, cited only for the purpose of information) the subject matter of **claim 1**, as far as examined (c.f. item III), does not appear to involve an inventive step. The skilled person is aware of the possible immunogenicity or toxicity of e.g. humanized Ab (as also recognized by the applicant in his letter of 07.06.2004, on page 3, line 23-24) and the different modern techniques suitable for the provision of e.g. human Ab (c.f. **D2** or description of the present application page 66, example 1) and, thus, would not need inventive skills to produce such a human Ab.

Notwithstanding, if the applicants argumentation (c.f. letter of 07.06.2004) would apply, i.e. that studies of humanized Ab have consistently shown minimal or no immune response in humans as exemplified e.g. by those already on the market, and, thus, the skilled person would have no incentive to produce a human Ab, the subject matter of **claim 1** would still appear to lack an inventive step as the invention would consist merely in choosing from a number of equally likely alternatives, i.e. humanized versus human anti-Ox40-Ab.

Therefore, the subject matter of **claim 1**, as far as examined (c.f. item III), does not appear to involve an inventive step in the sense of **Article 33(3), PCT**.

Independent **claims 5, 6-9, 11-17, 19-27**, as far as examined (c.f. item III), referring to functional variants of/immunoconjugates comprising/nucleic acids encoding/vectors comprising/hosts comprising/methods of producing/methods of identifying or obtaining human anti-Ox40 Ab by phage display technology as well as compositions comprising said molecules and their use as medicaments in general and more specifically for treating e.g. cancer (c.f. item III) do not contain subject matter which would require inventive skills from the skilled person being aware of the teachings in **D1** and that e.g. the phage display technology is a standard technology in the field of Ab production (c.f. **D2** or description of the present application page 66, example 1).

Also the subject matter of dependent **claims 3, 4, 10, 18, 28** does not add subject matter which would involve an inventive step in the sense of **Article 33(3), PCT**.

Thus, the subject matter of **claims 1, 3-28**, as far as examined (c.f. item III), does not appear to involve an inventive step in the sense of **Article 33(3), PCT**.

3.2 claim 2

The difference between **D1** and the subject matter of **claim 2**, as far as examined (c.f. item III), is the disclosure of human agonistic binding Ab capable of binding to and stimulating the human OX40 receptor and having synergistic stimulatory effects when co-incubated with OX-40L.

The problem to be solved is to provide agonistic binding Ab capable of binding to and stimulating the human OX40 receptor with no intrinsic immunogenicity or toxicity and having synergistic stimulatory effects when co-incubated with OX-40L.

The claimed solution is the use of human agonistic binding Ab as disclosed e.g. in example 7 of the present application (c.f. scFv 02-008 and 02-023 = SeqID No 25 and 28; page 76, §1 and Fig. 14A and B).

Starting from the closest prior art, the skilled person would have no incentive that he could obtain a human anti-Ox40 Ab with synergistic stimulatory effect when co-incubated with Ox-40L.

Therefore, the subject matter of **claim 2**, as far as examined (c.f. item III), would appear to involve an inventive step in the sense of **Article 33(3), PCT.**

4 Further remarks

4.1 Claim 24 is unclear in the sense of **Article 6, PCT** as far as relating to a disease.

4.2 Vague statements in the description such as on page 66, §2 imply that the subject-matter for which protection is sought may be different to that defined by the claims, thereby resulting in lack of clarity (**Article 6 PCT**) when used to interpret them (see also the **PCT Guidelines, III-4.3a**).

4.3 In a later European regional phase objections might be raised against any expression within the description such as "...which are incorporated herein in their entirety..." (such as on page 66, example 1, §2; page 67; line 6-7) as the regional patent law requires that the application is self-contained.

CLAIMS

1. An agonistic binding molecule capable of binding to and stimulating the human OX40-receptor.

2. A binding molecule according to claim 1, wherein the binding molecule is a human binding molecule.

3. A binding molecule according to claim 1 or 2, wherein the binding molecule comprises at least a CDR3 region comprising the amino acid sequence selected from the group consisting of SEQ ID NO:17 (DRYSQVHYALDY), SEQ ID NO:18 (DRYVNTSNAFDY), SEQ ID NO:19 (DMSGFHEFDY),SEQ ID NO:20 (DRYFRQQNAFDY),SEQ ID NO:21 (ARAAGTIFDY),SEQ ID NO:22 (DRYITLPNALDY),SEQ ID NO:23 (YDEPLTIYWFDS) and SEQ ID NO:24 (YDNVMGLYWFDY).

4. A binding molecule according to any one of the claims 1 - 3, wherein the binding molecule comprises a heavy chain comprising an amino acid sequence selected from the group consisting of SEQ ID NO:25, SEQ ID NO:26, SEQ ID NO:27 and SEQ ID NO:28.

5. A functional variant of a binding molecule according to claim 3 or 4, wherein the functional variant is capable of competing for specifically binding to the human OX40-receptor.

6. An immunoconjugate comprising a binding molecule according to any one of the claims 1 - 4 or a functional variant according to claim 5, the immunoconjugate further comprising at least one tag.

7. A nucleic acid molecule encoding a binding molecule according to any one of the claims 1 - 4 or a functional variant according to claim 5.

8. A vector comprising at least one nucleic acid molecule according to claim 7.

9. A host comprising at least one vector according to claim 8.

10. A host according to claim 9, wherein the host is a cell derived from a human cell.

11. A method of producing a binding molecule according to any one of the claims 1 - 4 or a functional variant according to claim 5, wherein the method comprises the steps of:

a) culturing a host according to claim 9 or 10 under conditions conducive to the expression of the binding molecule or functional variant, and

b) optionally recovering the expressed binding molecule or functional variant.

12. A binding molecule or functional variant thereof as obtainable by the method according to claim 11.

13. A method of identifying a binding molecule specifically binding to the human OX40-receptor or a nucleic acid molecule encoding a binding molecule specifically binding to the human OX40-receptor, wherein the method comprises the steps of:

a) contacting a phage library of binding molecules with material comprising the human OX40-receptor,

b) selecting at least once for a phage binding to the material comprising the human OX40-receptor, and

c) separating and recovering the phage binding to the material comprising the human OX40-receptor.

14. A method of obtaining a binding molecule specifically binding to the human OX40-receptor or a nucleic acid molecule encoding a human binding molecule specifically binding to the human OX40-receptor, wherein the method comprises the steps of:

a) performing the method according to claim 13, and

b) isolating from the recovered phage the binding molecule and/or the nucleic acid molecule encoding the binding molecule.

15. A composition comprising a binding molecule according to any one of the claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, or a binding molecule or functional variant thereof according to claim 12.

16. A composition comprising a nucleic acid molecule according to claim 7.

17. A pharmaceutical composition comprising a binding molecule according to any one of the claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a binding molecule or functional variant thereof according to claim 12, or a composition according to claim 15 or 16,

the pharmaceutical composition further comprising at least one pharmaceutically acceptable excipient.

18. A pharmaceutical composition according to claim 17 further comprising at least one other therapeutic agent.

19. Use of a binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for stimulating T-cells *in vitro*.

20. A binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for use as a medicament.

21. A binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for use in the treatment of a neoplastic, viral or bacterial disorder or disease.

22. A binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for use in enhancing the immune response in a human or animal.

23. A binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for use in enhancing the immune response against a tumour, bacterial or viral antigen in a human or animal.

24. Use of a binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for the preparation of a medicament for the treatment of a neoplastic, viral or bacterial disorder or a disease.

25. Use of a binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for the preparation of a medicament for enhancing the immune response in a human or animal.

26. Use of a binding molecule according to any one of claims 1 - 4, a functional variant according to claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a binding molecule or functional variant thereof according to claim 12, a composition according to claim 15 or 16 or a pharmaceutical composition according to claim 17 or 18 for the preparation of a medicament for enhancing the immune response against a tumour, bacterial or viral antigen in a human or animal.

27. A method for modulating a T-cell response in a human, comprising the step of administering to said human an effective dose of a binding molecule according to any one of the claims 1 - 4 or a functional variant of claim 5, an immunoconjugate according to claim 6, a nucleic acid molecule according to claim 7, a vector according to claim 8 or a pharmaceutical composition according to claim 17 or 18.

28. A method according to claim 27, wherein said modulation comprises the stimulation of T-cell proliferation.